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BIOLOGICAL EVALUATION R10-90-1

PATHOGENICITY OF *PHOMA* SP. ISOLATED FROM SEED OF LARCH SPECIES IMPORTED FROM THE SOVIET UNION

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Prepared by: Paul Hennon

Paul E. Hennon, Plant Pathologist
Forest Pest Management
Region 10, Alaska

Approved by: Gene Lessard

Gene Lessard, Group Leader
Forest Pest Management
State and Private Forestry
Region 10, Alaska

James E. Eggleston

James E. Eggleston, Director
Forest Pest Management
State and Private Forestry
Region 10, Alaska

Forest Pest Management
State and Private Forestry
Alaska Region
USDA Forest Service
P.O. Box 21628
Juneau, Alaska 99802

ABSTRACT

Phoma sp., isolated from seeds of larch imported from the Soviet Union, was tested for pathogenicity to determine if it is a threat to native larch or other vegetation in Alaska. The fungus lacked pathogenicity in spore suspension sprays and stem inoculations of four larch species, except that necrotic lesions developed in several stem inoculations on *L. decidua*. Three other species of larch, including *L. laricina* which is native in Alaska, were unaffected by inoculations. The fungus was tentatively indentified as *P. glomerata* and is probably not a threat to native larch or other vegetation in Alaska. It is known primarily as a saprophyte and is already extensively distributed throughout the world.

INTRODUCTION

Species of larch (*Larix* Mill.) that are native to the Soviet Union have the potential of being commercially important timber producing trees or ornamental trees if grown in Alaska (J. Alden, Personal communication). They are reportedly fast-growers, cold-tolerant, and well adapted to continental and maritime climates and a variety of soil types. These exotic larches may have a faster height and radial growth than the single species of native larch in Alaska, *L. laricina* (Du Roi) K. Koch.

Dr. John Alden, research forest geneticist with USDA Forest Service in Fairbanks, Alaska, received seeds of *L. sibirica* Ledeb. and *L. sukaczweii* N. Dyl. from the Moscow Region of the Soviet Union. Dr. Alden is interested in conducting studies on how these larches establish and perform as planted seedlings in Alaska. An examination of these seeds by Animal and Plant Health Inspection Service (APHIS) of USDA, however, resulted in the quarantining of these seeds because of the presence of a species of *Phoma*, an asexual fungus. Dr. Alden requested us (Forest Pest Management, R-10) to evaluate the occurrence of *Phoma* in seeds of these two Larch species and its pathogenicity on larch.

In a previous report (Hennon 1988), I determined that 2% of imported *L. sibirica* seeds were infected with a species of *Phoma* but seeds of *L. sukaczewii* were not infected. Based on gross cultural characteristics and microscopic structures *in vitro*, the fungus closely resembles *Phoma glomerata*. In this report, I evaluate the pathogenicity of the fungus on several species of larch, including *L. laricina*, to determine if the fungus is a pathogen or poses any threat to native larch in Alaska.

MATERIALS AND METHODS

Seedlings. Pathogenicity on larch was determined by wound inoculations on stems and by spraying spore suspensions on foliage and twigs of seedlings of *L. laricina*, *L. sibirica*, *L. sukaczweii*, and *L. dahurica* Trucz. The latter species, sometimes referred to as *L. gmelinii* (Rupr.) Lit., is native to eastern Siberia in the Soviet Union. Inoculated seedlings were grown in 10 cubic inch Ray-Leach cells and were either one or two years old. Seedling basal diameters ranged from 5 to 9mm. All inoculations were conducted on August 15, 1988 at the USDA Forest Service nursery at Petersburg, Alaska. Seedlings were evaluated for disease on June 8, 1989.

Stem-Wound Inoculations. Small wounds (2mm wide x 3mm long) were made on the lower stem of seedlings about 2cm above the soil line. A 6mm x 8mm piece of inoculum was cut from a live culture of *Phoma* and placed against each wound. The inoculum was wrapped against the wound with moist sterile cheesecloth and was then secured with 2.5 cm wide waterproof tape. Three inoculation treatments were randomly assigned to 15 seedlings for each of the four larch species. Treatments consisted of two different isolates of *Phoma* which originated from seeds of *L. sibirica* and, for control, sterile medium without fungus. Thus, for each of the four larch species, 30 seedlings received inoculation by *Phoma* and 15 seedlings received control-inoculation.

All seedlings were evaluated for infection with *Phoma* by unwrapping the wound and scrapping the outer bark away to examine for necrotic lesions or discoloration of the phloem or sapwood. Isolation was attempted from any symptomatic tissues that developed.

Spore suspension inoculations. A suspension of *Phoma* spores was made by flushing sterile water across cultures of *Phoma* that contained sporulating pycnidia. Suspensions were diluted to a concentration of 1,100,000 spores/ml. Viability of spores was confirmed by spraying the dilute solution onto fresh medium and observing growth of the fungus. Wounds caused by clipping three shoots from the of top each seedling and numerous leaf scars provided potential infection courts for the fungus. The dilute spore suspension was sprayed to runoff onto the entire above-ground portion of seedlings. An estimated 6,000,000 spores were sprayed onto each seedling. Sterile water sprayed in the same manner other seedlings served as a control. The number of seedlings sprayed for each larch species ranged from 13 to 16.

Seedlings were evaluated for infection with *Phoma* by examining needles and twigs for necrotic areas. Isolation of any symptomatic tissues was attempted.

RESULTS AND DISCUSSION

No seedling died from inoculation. Three seedlings, all *L. dahurica*, developed necrotic lesions from stem inoculations with *Phoma*. The three lesions measured from 16 to 19mm long and 4 to 7mm wide. All three were callusing shut and were no longer expanding, but *Phoma* was re-isolated from one of the lesions in June, 1989. Inoculation wounds on all other seedlings were callused shut and had not developed lesions.

Inoculation by spore suspension caused no injury to seedlings. Small dead twigs were found on three seedlings, but two were controls. *Phoma* was not isolated from any dead twigs.

Phoma showed no capability for inciting disease on native larch in Alaska. Weak pathogenicity was limited to *L. dahurica* and these seedlings did not die.

Of the more than 2,000 species of *Phoma*, most are saprophytes. The pathogenic species of *Phoma* typically cause disease of seedlings. *P. glomerata*, the probable identity of the fungus on seeds of *L. sibirica* (Hennon 1988), is not known as a pathogen of plants; it is ubiquitous on a wide variety of substrates (Domsch et al. 1980, Dorenbosch 1970, Sutton 1980). When it has been found in association with dead diseased material, the fungus is known as a secondary invader (Dorenbosch 1970). The species has been previously

reported from the U.S. and Canada and much of the world (Sutton 1980), but not from Alaska. It may already be present in Alaska and not reported due to the lack of mycological study on imperfect fungi in this region.

It now appears that the fungus present on *L. sibirica* seeds is not a threat to the native *L. laricina* in Alaska. Based on the results of this study, there is little reason to continue the quarantine on seeds of *L. sibirica* and *L. sukaczweii* imported from the Soviet Union.

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